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Laboratory cultures of Gambierdiscus toxicus were established with clones from wild populations in Hawaii and Tahiti French Polynesia. Seasonal variation of G. toxicus in Hawaii was monitored over a 16 month period. Influence of phosphate concentration on G. toxicus yield and toxicity was studied. Ciguatoxin (160 µg) was isolated from viscera of toxic eels (Gymnothorax javanicus) procured from Tarawa atoll, Republic of Kiribati.

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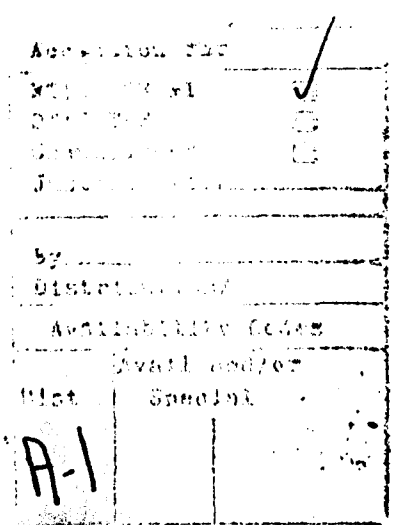

TABLE OF CONTENTS

I. INTRODUCTION

A. The Problem

Marine biotoxins are among the most potent naturally occurring toxins known. Their physiological actions are diverse, as are their molecular structures, not all of which have been fully determined. Some of these toxins constitute a hazard to human health and safety. It is therefore imperative to elucidate their structures and to make pure toxins available for the study of pharmacological properties and mechanisms of action.

In this study we are principally concerned with two toxins that are associated with the human fish intoxication known as ciguatera, ciguatoxin and maitotoxin, of unknown structure at the outset of this research, and with palytoxin, the structure of which is known but unique and whose mechanism of action is not well understood. The gross structure of ciguatoxin became known while this work was in progress. Each of the three toxins presents a different set of problems.

1. Ciguatoxin

It is present in ciguateric carnivorous fishes in concentrations ranging from 1 to 10 ppb. Its molecular structure has been elucidated. It has been isolated from toxic fish extracts and a closely related toxin from wild populations of the dinoflagellate Gambierdiscus toxicus, which is associated with ciguatoxic coral reefs.[1,2] It has not so far been recovered from laboratory cultures of G. toxicus.

2. Maitotoxin

In contrast to ciguatoxin, which is soluble in organic solvents, maitotoxin is water-soluble. It was first described from the gut of herbivorous toxic fishes, where it occurs in low concentration. It has been produced in a number of laboratories from cultured G. toxicus. Its molecular structure is partially known.

3. Palytoxin

This toxin, whose structure has been known for several years, has so far been isolated from toxic zoanthid corals, Palythoa spp. Preliminary evidence suggests that the producer of this toxin is not the coral, but an epiphyte, possibly a bacterium.

B. Previous Work

1. Ciguatoxin

There is no convincing literature report that would indicate that anyone has yet succeeded in producing ciguatoxin from cultures of G. toxicus. Miller and coworkers[3] have cultured G. toxicus from a Caribbean collection. The nature of the toxin is difficult to assess as these workers report an LD_{50} of 4.95 mg/kg of the lipid extract after four chromatographic purification steps. This corresponds to but 0.01% of the lethality of pure ciguatoxin. Durant-Clement[4] has cultured G. toxicus from a Gambier islands strain and reports production of toxic extracts, water-soluble maitotoxin-like and lipid-soluble ciguatoxin-like, in ratios varying from 9:1 to 7.5:2.5.

The bulk of the early work on the molecular structure of ciguatoxin, was performed by Tachibana.[5] Yasumoto and coworkers[1,2] successfully concluded the structural elucidation, except for stereochemical detail.

2. Maitotoxin

Maitotoxin has been produced from G. toxicus cultures in a number of laboratories. Yasumoto and coworkers[6] have had G. toxicus from the Gambier islands in culture since soon after the organisms was described.[7] They have been producing maitotoxin consistently and have begun mass spectral and NMR studies to delineate its structure.

3. Palytoxin

The molecular structure of palytoxin is known,[8] but the suggestion [9] that the toxin is biosynthesized by an epiphyte of the zoanthid coaral has not been realized.

C. Purpose of Present Work

The purposes of this work are the successful culture of G. toxicus; production of ciguatoxin and maitotoxin; structural elucidation of both toxins; and production of palytoxin in laboratory culture.

D. Method of Approach

1. G. toxicus culture

The methodology is known.[3,4,6] What is not known, however, are the factors, genetic and/or environmental, which govern production of maitotoxin and/or ciguatoxin. Hence the approach must be empirical. We will collect G. toxicus from as many sites as possible and experiment with a variety of culture parameters.

2. The Molecular Structures of Maitotoxin and Ciguatoxin

Once the G. toxicus cultures are productive, they presumably will furnish maitotoxin. This toxin can be purified and its structure elucidated by well-known procedures and techniques.

Until such time, when the G. toxicus cultures produce ciguatoxin, we will continue to procure toxic carniverous fishes and purify ciguatoxin for structural elucidation. Because of the extremely low concentration of toxin our goal will be the production of a crystalline derivative for structure determination by x-ray diffraction techniques.

3. Palytoxin Production from Cultures

This phase of the research was to be initiated once the toxin production from G. toxicus cultures had been achieved. However, the contract was terminated prematurely.

II. RESULTS AND DISCUSSION

Because of initial administrative delays in activating this contract and because of constructing and equipping a new culture laboratory, the effective starting date of the research was delayed by approximately six months.

A. Equipment for the Culture Laboratory

This contract provided funds for equipping a newly constructed laboratory. Major acquisitions are listed in Table I.

B. G. toxicus Cultures

1. Dinoflagellate sources

G. toxicus culture samples were requested from Drs. Sherwood Hall (USFDA), R. Bagnis (French Polynesia), and Jeffrey Bomber (SIU, Carbondale), but none were received. Consequently, G. toxicus collections were made from wild populations on the island of O'ahu on a continuing basis. One field collection was made in Tahiti, French Polynesia. The consistently most favorable site on O'ahu proved to be Kahala Beach (site 4, Fig. 1). Results are documented in Fig. 1 and Table II. The Kahala Beach site was monitored from September, 1988, through December, 1989, for population density of G. toxicus (•) and Ostreopsis siamensis (▬). Both organisms showed substantial increase over the 16 month study period. However, the cell density is far below values recorded from French Polynesia. The results are shown in Fig. 2.

2. Dinoflagellate Culture

Cultured cells were harvested and sequentially extracted with acetone (A) and methanol (M). Lethality of the extracts was evaluated by mouse bioassay. The results are listed in Table III.

3. Effect of Phosphate Concentration on G. toxicus culture

Because of the unspectacular results of the culturing experiments, a systematic evaluation of environmental parameters was undertaken. The first series of experiments dealt with the effect of phosphate concentration on yield and toxicity of cultured G. toxicus.

a. Methodology

Six different concentrations of phosphate were used: 0 M, 1 x 10 -4 M, 5 x 10 -5 M, 1 x 10 -5 M, 5 x 10 -6 M, 1 x 10 -6 M. All other constituents of the modified ES medium were unchanged. No attempts were made to remove phosphate if any in natural sea water. Clean, sterile 125 mL Erlenmeyer flasks containing 50 mL of medium were inoculated with 2500 cells (50 cells/mL. Each phosphate level was triplicated. Every five days, 1 mL of medium containing well-suspended cells were removed from each flask. The number of cells present and the condition of the cells were recorded. This was done by visually counting 7 random microscope views through a 10x lens on the inverted microscope. It was previously determined that the counting chamber was made up of 21 views at this power. The mean of the seven views was calculated, and this number was multiplied by 21 to obtain the daily count. The data are listed in Tables IV through IX. Each Table represents a different concentration of phosphate. The columns in each Table represent cell counts per mL for a given day. Each row represents a separate replicate experiment 1 through 6, followed by the triplicate number, 1 through 3. Each replicate lasted 35 days, when the cells were harvested. A total of 6 replicates were conducted. A cumulative Table (Table X) contains the mean count values for each concentration. A graphical view of the data is illustated in Fig. 3. Each concentration is represented by a separate curve. Each curve is superimposed on the same graph for comparison purposes.

When the experiments were terminated after 35 days, the cells from each Erlenmeyer flask of one concentratrion were pooled and harvested onto a glass microfiber filter. The filter and a vial were pre-weighed. After the cells were harvested, they were allowed to air-dry on an aspirator for 5 minutes.

After 5 minutes, the glass microfiber filter was returned to the vial and weighed again. The data are shown in tables XI through XVI. The vials are numbered arbitrarily; the assigned number can be found in the "Expt" column. The first number represents the vial from which the replicate originated. Please note that a 0 M phosphate concentration was not conducted in replicate 1. Afterwards the cells were extracted to evaluate the toxicity.

In order to extract the cells with maximum efficiency, the cells were extracted directly from the filter. Approximately 10 mL of methanol was placed in each vial. The vials were then placed in an ice bath in a sonicator. Each vial was sonicated for 30 minutes. The solvent was then decanted through Whatman fluted filter paper and fresh methanol was placed in the vial. This solvent remained in the vial for 24 hours. After 24 hours, the vial was again sonicated for 30 minutes. The solvent was decanted and the process was repeated once more. The entire extraction procedure was carried out over 48 hours; methanol in the vials was changed and the vials were sonicated 3 times. The solvent from each of the 3 extractions were combined for each concentration. The concentrations were kept separate by replicates. A total of 35 vials of methanol extract were screened for toxicity. Each vial containing methanol extract was submitted for cytotoxic testing against KB cells. The concentration submitted was 1 mg/mL dissolved in a 1:1 ethanol : water solution. The cytotoxicity results are also recorded in Tables XI through XVI.

b. Results may be seen in Fig. 4, the maximum average growth rate was achieved at a phosphate concentration of 5 x 10 -5 M. The general trend as illustrated in the Figure shows a decline in the growth rate as phosphate concentration decreases. The Figure also shows that increased phosphate concentration above 5 x 10 -5 M does not promote an increase in growth rate. It can be safely concluded that increased phosphate will increase growth till a maximum is reached. The concentration used as the control in this experiment was 1 x 10 -5 M. However, the two experiments did not show large differences; therefore, a five-fold variation may not be significant. In fact, all concentrations other than no phosphate and 1 x 10 -6 M phosphate exhibited similar growth curves. As for toxicity trends, the results obtained were inconclusive. It may have been better if the mouse bioassay has been used. Lack of time and limited amounts of extracts precluded this.

C. Molecular Structure of Ciguatoxin

Two sources of toxic fish were used for the isolation and purification of ciguatoxin. Approximately 500 kg of Seriola dumerilii (yellowtail amberjack), many of which had tested positive in the Hokama stick test was donated to us from commercial fishermen. The fish had originated in the Northwest Hawaiian islands (Nihoa to Midway). This source, although unfit for human consumption, proved to be unrewarding for ciguatoxin isolation.

Once again, we established connection with Tarawa atoll, Republic of Kiribati, for the procurement of moray eel (Gymnothorax javanicus) viscera. From 21.75 kg of viscera we isolated 160 μg of ciguatoxin at a stage prior to final HPLC purification.

When the gross molecular structure of ciguatoxin (Fig. 4) became known,[1,2] it was apparent that a terminal vic 1,2-diol was the characteristic structural entity of the toxin. Hence it seemed desirable to shift emphasis toward utilizing this structural feature (1) for the production of derivatives suitable for x-ray diffraction in order to solve the stereochemistry of the molecule; (2) for fluorescence detection during chromatography; and (3) for protein conjugation and ultimate antibody production.

None of these experiments had reached a conclusive stage, when the contract was prematurely terminated.

D. Palytoxin

No work had been initiated, when the contract was terminated.

III. CONCLUSIONS

The following problems remain to be solved.

A. G. toxicus Culture

1. Parameters, genetic and/or environmental, which govern toxin production of G. toxicus.
2. Molecular structure of maitotoxin.
3. Stereochemistry of ciguatoxin.
4. Production of a ciguatoxin derivative suitable for protein conjugation
5. Which organism produces palytoxin?

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Table I. Major Equipment Acquisition

Fermenters, 5 and 30 L working volume

Computer control system for above

Chiller for 30 L fermenter

Controlled rate freezer

Cryogenic container

Backup air compressor

Inverted Microscope

Backup generator

Low temperature freezer

Controlled environment shaker

Media Preparation equipment

Centrifuge rotor

Tissue homogenizer

Refrigerated recirculator for 5 L fermenter

Alarm system

Table II. Species of Macroalgae from Sites on the Island of O'ahu

ALGAL SPECIES	SITE	SUBSTRATE	PRESENCE OF	
			Gambierdiscus toxicus	*Ostreopsis siamensis*
Chlorophyta.				
Halimeda opuntia	4	sand	-	-
H. discoidea	4	sand	-	-
Enteromorpha sp.	7	mud	-	-
Phaeophyta				
Turbinaria ornata	4	rock	-	-
Sargassum polyphyllum	4	rock	-	-
S. echinocarpum	3	rock	-	-
Padina sp. 1	4	rock	+	-
Padina sp. 2	5	rock	-	-
Dictyota dichotoma	4	sand	-	-
D. sandvicensia	4	sand	+	+
D. acutiloba	4	sand	-	-
Rhodophyta				
Spyridia filamentosa	1	rock	-	-
	3	sand/ rubble	+	+
	4	sand/ rubble	+	+
	5	sand/mud	+	+
	6	sand	-	-
	7	mud	-	-
	8	sand	+	+
Acanthophora spicifera	2	mud	-	+
	4	sand/ rubble	+	+
Plocamium sandvicense	3	rock	-	-
Laurencia obtusa	4	rock	-	-
L. succisa	4	rock	-	-
Grateloupia filicina	3	sand	-	-
UNIDENTIFIED ALGAE				
Brown filamentous 1	8	sand	-	-
Brown filamentous 2	7	mud	-	-
Green filamentous	7	mud	-	-

Table III. Cell and Extract Yields, Toxicity of Mice of Crude Extracts from Cultured Dinoflagellates

	YIELD OF CELLS (mg)	YIELD OF EXTRACT (mg) A	YIELD OF EXTRACT (mg) M	TOXICITY TO MICE A	TOXICITY TO MICE M
BATCH CULTURE					
GT; KB					
(A)	23.4	13.0	24.0	+	+(L)
(B)	35.6	1.6	5.0	+	+(L)
GT: Tahiti	22.2	13.0	13.4	+	+(L)
CLONAL CULTURES					
GT 174	242.2	22.0	14.0		
GT 177	269.9	12.0	16.0	+	+(L)
PL 4 (A)	90.3	41.3	7.9	-	+(L)
PL 4 (B)	108.3	12.1	6.4	-	+(L)

GT - Gambierdiscus toxicus

PL - Prorocentrum lima

KB - Kahala Beach

L - lethal

\+ - toxic symptoms

Table IV. Data for (PO4)=0

Day	5	10	15	20	25	30	35
1-1							
1-2							
1-3							
2-1	109	210	209	223	237	219	273
2-2	73	158	247	254	285	225	201
2-3	52	150	255	263	231	227	215
3-1	71	120	169	170	175	255	165
3-2	74	149	207	170	210	190	165
3-3	54	107	163	245	165	170	150
4-1	102	88	102	145	145	160	215
4-2	45	87	130	210	155	130	160
4-3	43	69	107	165	180	160	155
5-1	58	115	155	170	168	177	162
5-2	42	119	155	150	270	147	158
5-3	45	97	245	175	195	228	198
6-1	182	222	270	214	228	240	252
6-2	194	262	200	191	177	201	195
6-3	187	220	330	239	288	198	156
mean	89	144	196	199	207	195	188

Table V. Data for (PO4)=1x10 -6

Day	5	10	15	20	25	30	35
1-1	55	108	292	530	600	1020	605
1-2	76	148	343	465	430	535	490
1-3	97	63	136	35	485	540	700
2-1	66	164	305	480	340	385	380
2-2	92	158	342	585	435	345	380
2-3	86	179	383	605	410	280	445
3-1	42	65	142	235	280	305	335
3-2	40	53	95	190	415	355	230
3-3	47	70	124	260	350	255	495
4-1	94	107	253	220	315	395	345
4-2	35	84	177	345	300	295	415
4-3	38	118	296	285	330	395	340
5-1	90	269	490	500	450	630	339
5-2	70	182	470	620	360	558	405
5-3	105	243	425	485	471	558	504
6-1	192	555	580	595	558	537	501
6-2	119	405	385	840	507	393	591
6-3	163	345	650	850	528	555	621
mean	83	184	327	450	420	463	451

Table VI. Data for (PO4)=5x10 -6

Day	5	10	15	20	25	30	35
1-1	68	200	382	695	1175	1175	1500
1-2	97	181	449	1150	1650	1525	1485
1-3	62	88	268	730	1390	1205	1425
2-1	92	206	576	1040	1240	1570	1535
2-2	83	209	568	1110	1380	1360	1300
2-3	89	196	420	730	980	1115	1150
3-1	45	83	161	435	710	640	725
3-2	60	84	302	670	685	975	1165
3-3	64	77	192	550	600	740	1010
4-1	46	71	186	310	440	625	685
4-2	70	91	196	285	485	540	635
4-3	38	83	144	320	495	710	945
5-1	57	171	490	910	1233	2034	1515
5-2	93	296	645	1140	1530	1818	1986
5-3	66	235	585	1155	1755	1865	2031
6-1	161	585	980	1260	984	1320	1506
6-2	182	445	845	1245	1448	1683	1632
6-3	137	440	730	1215	1182	1530	2148
mean	84	208	452	831	1076	1246	1353

Table VII. Data for (PO4)=1x10 -5

Day	5	10	15	20	25	30	35
1-1	101	176	242	425	790	985	830
1-2	68	165	286	505	835	1005	1235
1-3	95	198	309	560	930	875	1040
2-1	74	157	478	765	950	1725	1835
2-2	69	189	567	900	1850	1835	1685
2-3	50	157	450	770	1420	1745	1375
3-1	63	113	303	500	1055	1205	1450
3-2	45	88	213	395	665	1260	1725
3-3	58	108	225	505	910	1185	1470
4-1	38	59	134	115	335	600	765
4-2	41	78	169	240	455	455	855
4-3	38	80	150	290	415	590	880
5-1	57	200	460	660	914	1062	1239
5-2	67	258	555	760	999	903	1164
5-3	64	165	345	595	693	843	1221
6-1	204	652	1205	1810	2331	2076	2703
6-2	182	489	1020	1520	2640	2316	2623
6-3	202	722	1200	1810	2667	2109	2124
mean	84	225	462	718	1159	1265	1457

Table VIII. Data for (PO4)=5x10 -5

Day	5	10	15	20	25	30	35
1-1	53	108	227	335	855	845	1125
1-2	117	150	310	650	865	1055	1350
1-3	74	90	240	500	719	1045	1310
2-1	43	134	344	530	920	1805	1430
2-2	100	250	565	900	1120	1415	1655
2-3	52	148	390	735	1090	1345	1810
3-1	70	105	294	680	1070	1380	2375
3-2	75	137	278	465	595	785	1020
3-3	64	107	2710	420	640	655	755
4-1	45	85	201	190	380	300	470
4-2	44	58	176	215	290	535	695
4-3	51	86	231	495	395	635	565
5-1	77	232	595	1055	1737	1611	1803
5-2	67	226	660	1115	1482	1863	2123
5-3	68	208	560	960	1548	2178	1605
6-1	175	720	1065	1710	1443	2289	2487
6-2	187	720	1295	1730	1758	2208	2643
6-3	242	79	1105	1760	1986	2328	2562
mean	89	202	625	802	1050	1349	1544

Table IX. Data for (PO4)=1x10 -4

Day	5	10	15	20	25	30	35
1-1	66	75	159	225	430	525	790
1-2	80	105	178	425	545	545	960
1-3	62	135	145	355	645	690	1005
2-1	60	113	305	455	805	890	1195
2-2	92	180	401	640	955	1005	1250
2-3	71	151	408	635	865	1125	1265
3-1	71	141	274	480	920	1160	1285
3-2	40	142	267	475	715	1045	1010
3-3	47	127	295	600	825	1350	885
4-1	44	59	115	185	215	470	600
4-2	49	58	139	305	220	745	805
4-3	63	68	157	205	360	505	750
5-1	79	127	550	820	1074	1503	1617
5-2	61	183	445	765	1098	1725	1899
5-3	78	188	365	625	1086	1209	1872
6-1	169	595	1380	1410	2289	2490	2679
6-2	176	686	1415	1265	2109	2709	2691
6-3	249	954	1340	1635	1494	2247	2511
mean	86	227	463	639	925	1219	1393

Table X. Mean values

0	1X10 -4	5X10 -5	1X10 -5	5X10 -6	1X10 -6	DAY
50	50	50	50	50	50	0
89	86	89	84	84	83	5
144	227	202	225	208	184	10
196	483	625	462	452	327	15
199	639	802	718	831	450	20
207	925	1050	1159	1076	420	25
195	1219	1349	1265	1246	463	30
188	1393	1544	1457	1353	451	35

Table XI. Data for (PO4)=0

Expt	wt(mg)	cytotoxicity
2-1	181.2	4+ 1ug/mL
3-1	153.3	3+ 10ug/mL
4-1	138.5	4+ 5ug/mL
5-1	203.5	3+ 1ug/mL
6-1	191.3	2+ 1ug/ml

Table XII. Data for (PO4)=1x10-6

Expt	wt(mg)	cytotoxicity
1-5	207.3	-
2-6	206.0	4+ 5ug/mL
3-6	178.2	4+ 10ug/mL
4-6	137.0	4+ 5ug/mL
5-6	310.1	4+ 1ug/mL
6-6	307.3	4+ 10ug/mL

Table XIII. Data for (PO4)=5x10-6

Expt	wt(mg)	cytotoxicity
1-4	253.9	2+ 5ug/mL
2-5	248.6	2+ 5ug/mL
3-5	154.6	-
4-5	138.8	4+ 1ug/mL
5-5	222.7	4+ 1ug/mL
6-5	239.8	4+ 1ug/mL

Table XIV. Data for (PO4)=1x10-5

Expt	wt(mg)	cytotoxicity
1-3	257.8	4+ 5ug/mL
2-4	241.7	2+ 5ug/mL
3-4	176.5	-
4-4	36.4	4+ 5ug/mL
5-4	314.4	4+ 1ug/mL
6-4	390.1	4+ 1ug/mL

Table XV. Data for (PO4)=5x10-5

Expt	wt(mg)	cytotoxicity
1-2	233.5	4+ 5ug/mL
2-3	243.4	4+ 1ug/mL
3-3	189.0	3+ 10ug/mL
4-3	191.5	4+ 1ug/mL
5-3	197.4	4+ 1ug/mL
6-3	283.0	3+ 1ug/mL

Table XVI. Data for (PO4)=1x10-4

Expt	wt(mg)	cytotoxicity
1-1	233.5	3+ 5ug/mL
2-2	243.4	4+ 1ug/mL
3-2	189.0	3+ 10ug/mL
4-2	191.5	-
5-2	197.4	4+ 1ug/mL
6-2	283.0	4+ 1ug/mL

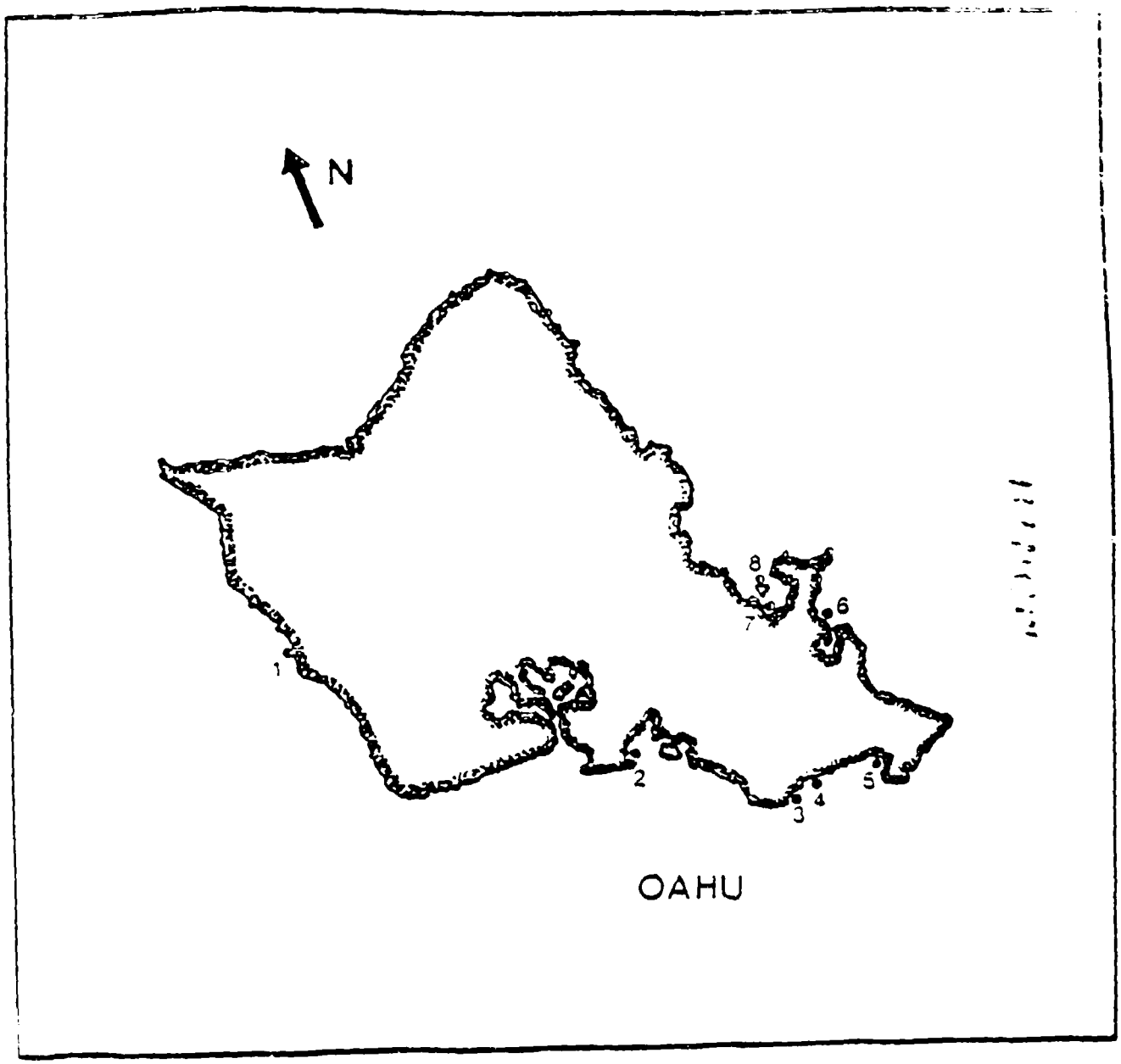
Figure Legends

Figure 1. A map of the island of O'ahu showing the location of collection sites: (1) Lualualei Beach Park (2) The reef runway, Keehi Lagoon side (3) Black Point (4) Kahala Beach (5) Hawaii Kai (6) Kailua Beach Park (7) Kaneohe Bay (8) Coconut Island.

Figure 2. Seasonal variation of G. toxicus (•) and Ostreopsis Siamensis (▪).

Figure 3. Effect of phosphate concentration on yield of cultured G. toxicus.

Figure 4. Molecular structure of ciguatoxin.


N
1
2
3
4
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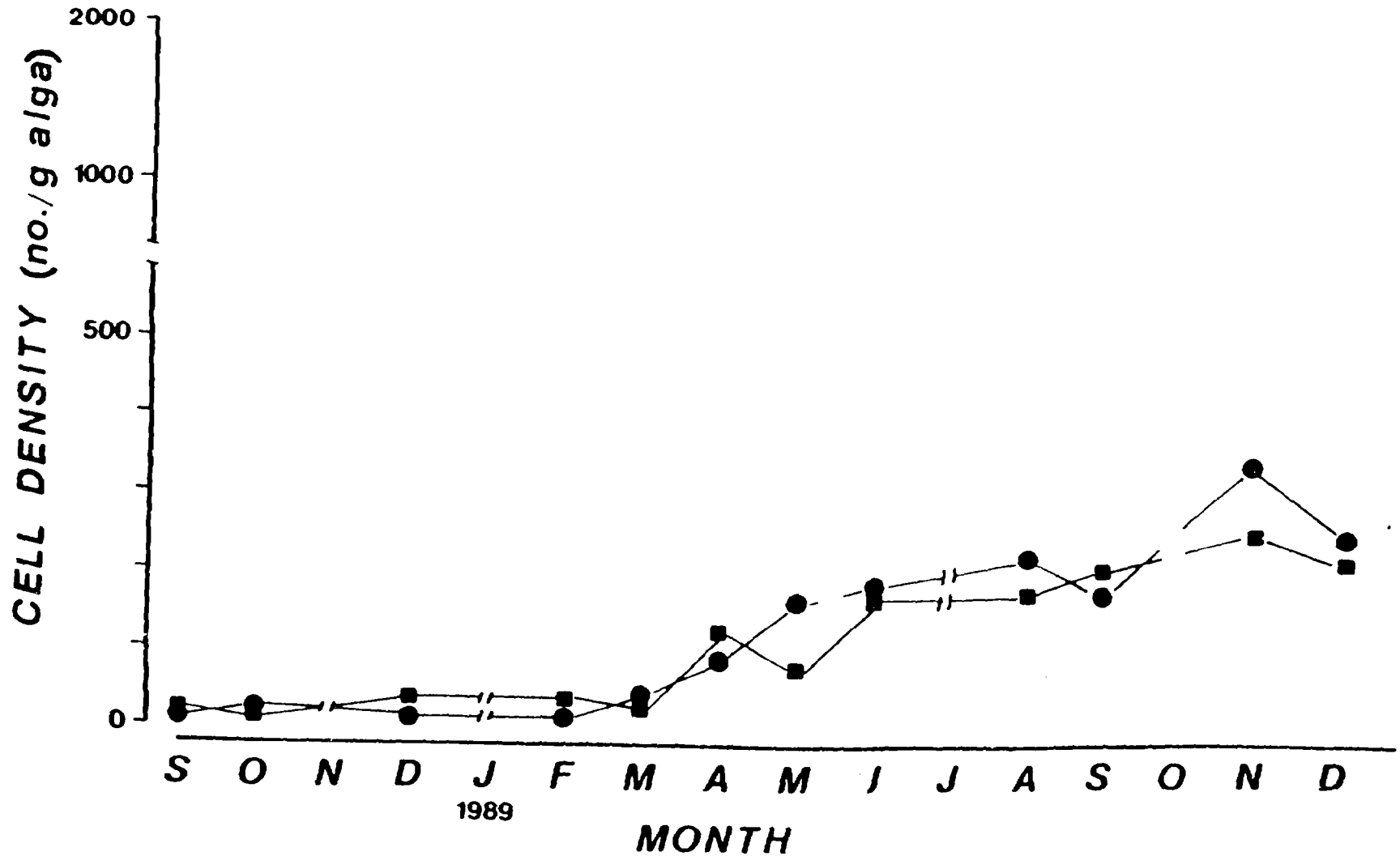
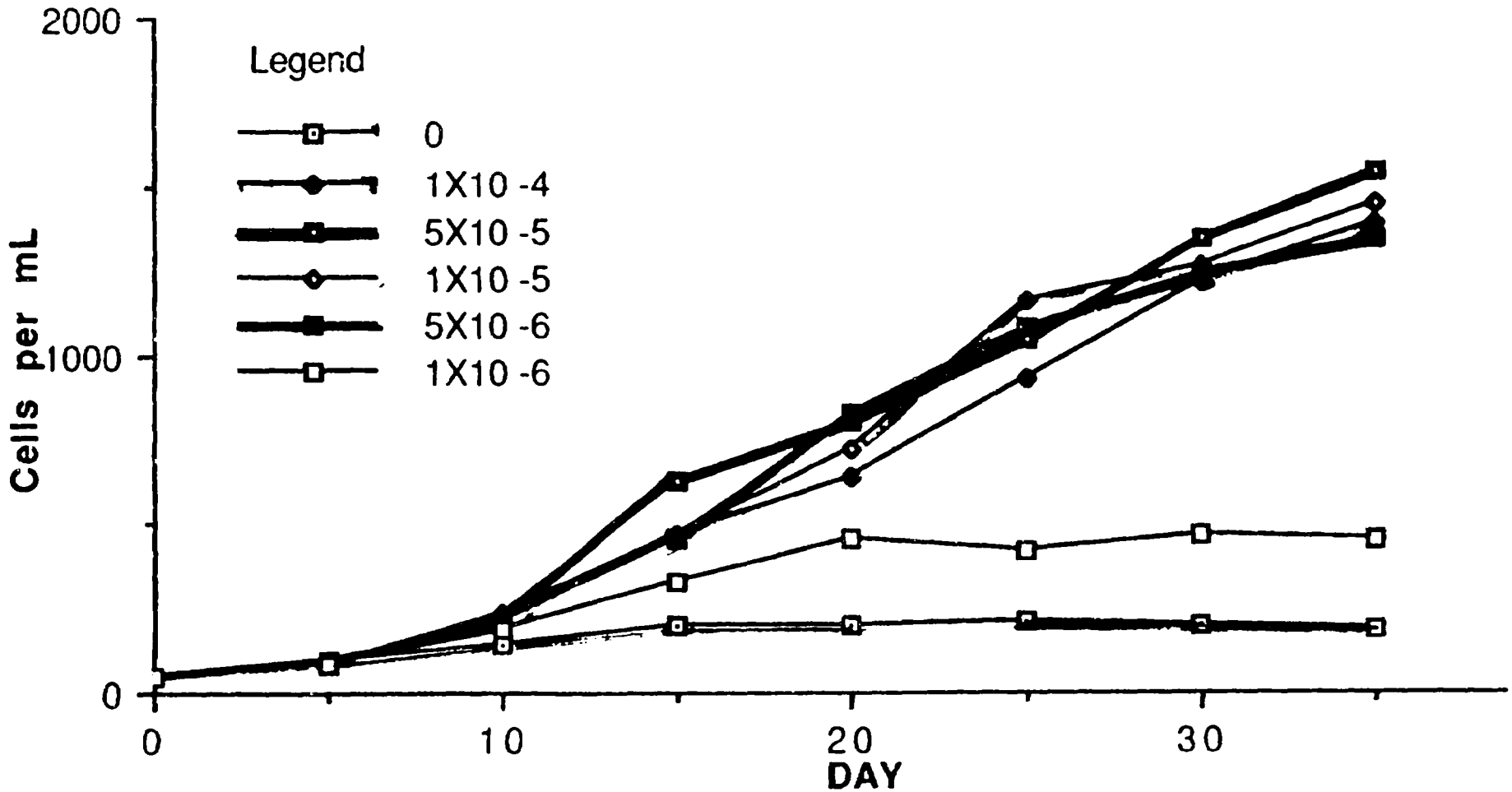
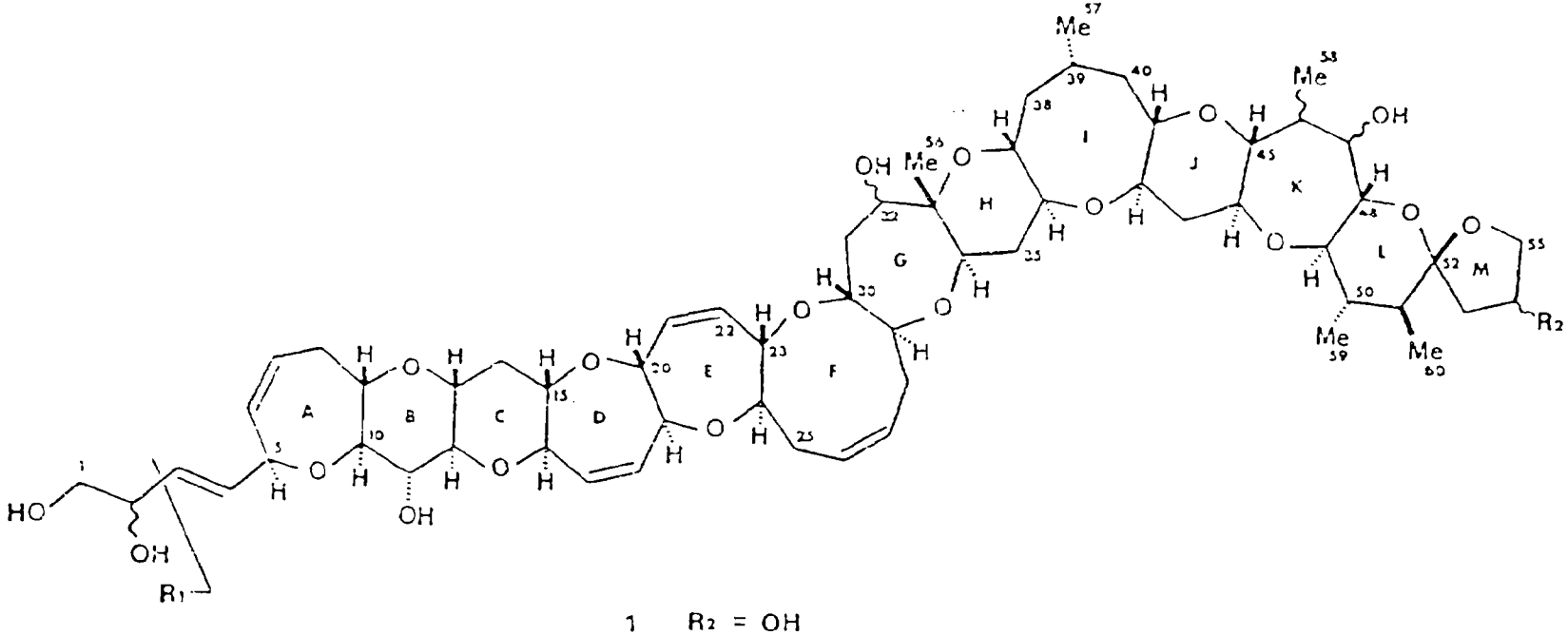
8
OAHU


2000
1000
500
0
CELL DENSITY (no./g alga)
S O N D J F M A M J J A S O N D
1989
MONTH


2000
1000
0
Cells per mL
Legend
0
1X10 -4
5X10 -5
1X10 -5
5X10 -6
1X10 -6
0
10
20
30
DAY



1 R_2 = OH

2 R_1 = CH_2^1=CH-
R_2 = H

STRUCTURES OF CTX ISOLATED FROM MORAY EEL (1) AND ITS CONGENER FROM THE CAUSATIVE DINOFLAGELLATE Gambierdiscus toxicus (2)

Contract No. DAMD 17-87-C-7210

Appendix

A. Bibliography

1. Ciguatera - what we know and what we would like to know.
P.J. Scheuer in "Mycotoxins and Phycotoxins '88", (S. Natori, K. Hashimoto, Y. Ueno, Eds.) Elsevier: Amsterdam, 1989, pp 265-276.

2. Seasonal Abundance and Toxicity of Gambierdiscus toxicus from O'ahu, Hawai'i.
E.J. McCaffrey, M.M.K. Shimizu, P.J. Scheuer, and J.T. Miyahara, in press.

B. Abstracts (attached)

1. 7th International IUPAC Symposium on Mycotoxins, and Phycotoxins, Tokyo, Japan, August 1988.

2. Third International Conference on Ciguatera, Puerto Rico, April 1990.

C. Personnel Receiving Pay
R.K. Akee
D.A. Corgiat
C.M. Matsumoto
E.J. McCaffrey
G.M.L. Patterson
S.G. Sakurai
M.M.K. Shimizu
P. Tong-On
W.Y. Yoshida

D. Graduate Degrees
None

1st International Toxic Symposium
on Mycotoxins and Phycotoxins

FORM D

Reg No.

Abstract Form for Speaker

CIGUATERA - WHAT WE KNOW AND WHAT WE WOULD LIKE TO KNOW

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Judging by the virtual torrent of recent conferences, symposia, and publications[1-7], one cannot escape the observation that there has been increasing awareness of the circumtropical public health problem that is referred to as ciguatera. The term denotes a human intoxication caused by the ingestion of tropical coral reef fish, or of biota that consume such fish. Yet despite the upsurge in the dissemination of existing knowledge, solutions to remaining questions and sophisticated understanding of this complex ecological phenomenon have not been forthcoming at a comparable pace.

In this lecture I will describe the major advances in ciguatera research beginning with Yasumoto's[8] decisive discovery in 1977 of the benthic dinoflagellate Gambierdiscus toxicus and highlight the unanswered questions in etiology, ecology, molecular structure, and detection.

References

1. Second International Conference on Ciguatera, San Juan, Puerto Rico, April 1985; see Marine Fish. Rev. 1986, 48 (4), 1-59.

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3. First International Workshop on Ciguatera, Beverly, MA, June 1985; see Biol. Bull. 1987, 172 (1), 89-153.

4. Nukina, M.; Tachibana, K.; Scheuer, P. J. In "Natural Toxins" (Harris, J. B., Ed.) Clarendon Press, Oxford, 1986, pp 46-55.

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8. Yasumoto, T.; Nakajima, I.; Bagnis, R.; Adachi, R. Bull. Jap. Soc. Sci. Fish. 1977, 43, 1021-1026.

Seasonal Abundance and Toxicity of Gambierdiscus toxicus from O'ahu, Hawai'i

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Inshore reef environments of the island of O'ahu were monitored for the presence of the benthic dinoflagellate Gambierdiscus toxicus from September, 1988 to December, 1989. Low population levels ranging from 15 cells/g alga to 225 cells/g alga were found in association with the macroalgae Spyridia filamentosa, Acanthophera spicifera, and Dictyota sandvicensis. S. filamentosa was the preferred substrate.

Several monoclonal and batch cultures of G. toxicus were isolated and grown in the laboratory. Lethality of crude acetone and methanol extracts were assayed by ip injection into mice. In vitro cytotoxicity and antiviral activity of these extracts was also evaluated.

Results of these studies will be discussed.

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*MBRS participant